Appendix
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APPENDIX 5.2.4.

DISINFECTION OF CRUSTACEAN FARMS

Article 5.2.4.1.

See Appendix 5.2.2 for general information on *disinfection*, commonly used *disinfectants*, recommendations for typical methods of application, working concentrations and target dose rates, and notes for their responsible use and disposal, especially as these practices relate to human and environmental safety.

Article 5.2.4.2.

General principles

The choice of a disinfection method for use in a crustacean farm depends on many factors, which may include: the reason(s) for *disinfection*, whether the 'farm' is a broodstock facility, hatchery, or growout farms; and the type of growout farm. Because the penaeid shrimp are the hosts for all but one of the crustacean *diseases* currently listed in the *Code*, this Appendix will be focused on penaeid shrimp.

Article 5.2.4.3.

Reason(s) for disinfection

Disinfection is employed as a common disease management tool in shrimp farming. It may be used as a routine practice in biosecurity programmes designed to exclude specific *diseases*, as well as a routine sanitary measure employed to reduce disease incidence within farms, or it may be used in disease eradication (stamping out) efforts. The specific reason for *disinfection*, will determine the disinfection strategy used and how it is applied.

Article 5.2.4.4.

Occurrence of listed diseases

When an OIE listed *disease*, or an important but unlisted *emerging disease* occurs for the first time at a particular farm, at a particular site (i.e. at a quarantine facility), or within a region or country previously believed to be free of that *disease*, it may be advisable, if not required, to eradicate the *disease* by depopulating the facility and performing a thorough *disinfection* of all or part of the facility. *Fallowing* of the affected facility for a defined period of time may be warranted in some situations (see Chapter 1.7.1, Fallowing of Sites in this *Code*).

Article 5.2.4.5.

Prevention of disease spread to wild populations

The direct disposal of diseased populations of live shrimp (any life stage; i.e. fertilised or unfertilised *eggs*, larvae, postlarvae, juveniles, or adults) or waste products derived from them (i.e. processing plant wastes such as shells, broken shrimp pieces, etc.) into receiving waters (i.e. creeks, rivers, estuaries, bays, littoral areas) is a dangerous practice that facilitates the spread of *disease* from farmed populations to wild crustacean stocks or to neighbouring farms that use the same water supply, and it should not be permitted to occur. With cultured stocks, when the decision is made to discard a population (i.e. that is being cultivated in a hatchery tank or growout pond) due to the presence of *disease* (or poor culture performance which may be due to an undiagnosed *disease*), the stock in the tank or pond should be harvested and/or humanely killed in the tank or pond. The water in the tank or pond should be disinfected (see specific sections on *disinfection* of tanks and growout ponds in the Article) prior to discharge. The emptied tank or pond should be disinfected prior to restocking.

Article 5.2.4.6.

Routine sanitation and biosecurity

Many crustacean farms, especially those cultivating penaeid shrimp, employ measures that use a number of disinfection methods for disease prevention and control. These measures may be part of a farm's routine biosecurity programme that may be designed for exclusion of specific *diseases* as well as serving as general pest and disease exclusion measures.

1. *Disinfectants*

   The following list comprises the *disinfectants* recommended for use in shrimp farms (the appropriate disinfectant regimes for each specific application are discussed in the appropriate sections):

   - chlorine (as calcium hypochlorite, HTH™ or a bleach solution that is 5.25% sodium hypochlorite);
   - formaldehyde gas (from sublimated paraformaldehyde or concentrated formalin/potassium permanganate reaction);
   - iodine (as contained in iodophors);
   - lime (as calcium oxide or calcium hydroxide);
   - UV light (from natural sunlight);
   - ozone;
   - steam;
   - hot water (60ºC);
   - concentrated acids;
   - desiccation;
   - detergents (for general cleaning, with some degree of disinfection capability for many products).

2. Hatcheries and broodstock rearing/holding facilities

Virtually all penaeid shrimp hatcheries and broodstock holding/rearing facilities use seawater that has been disinfected to remove potential pathogens, pests, and disease-carrying agents via mechanical filtration, UV irradiation, and/or chemical disinfection. This may be by passive source water filtration (i.e. by the use of seawater wells or well points) or by mechanical filtration using high pressure pumps and a variety of water filtration devices and pore sizes. Some facilities use filtration coupled with UV light disinfection of source water, while others use chemical disinfection methods, using either chlorination and de-chlorination or high doses of ozone and subsequent removal of residual oxidants. Chemical disinfection of source water typically requires the use of one or more water storage reservoirs in which the water is treated and detoxified before use in the shrimp hatchery or broodstock facility. Numerous manuals are available that provide specifics on hatchery and broodstock facility design and operation for shrimp culture, and in which details on source water disinfection are provided.

a) *Disinfection* of *eggs* and larvae in penaeid shrimp hatcheries

Certain penaeid shrimp viral *diseases* (i.e. Spherical baculovirosis, Tetrahedral baculovirosis, Hepatopancreatic parvovirus infections) are transmitted by faecal contamination of spawned eggs. These *diseases*, as well as infections due to certain other shrimp viruses such as White spot disease virus, and certain bacterial and fungal *disease agents*, can be eliminated or have their incidence reduced through the routine use of disinfection protocols when used to surface disinfect eggs and/or recently hatched nauplii. A widely used method is given below:

For fertilised eggs[1]

Collect fertilised eggs. Rinse with running seawater for 1–2 minutes. Dip in 100 ppm (parts per million) formalin for 1 minute. Dip in iodophor (0.1 ppm iodine) for 1 minute. Rinse in running seawater 3–5 minutes. Transfer to disinfected larval rearing tanks.

For nauplii[2]

Using phototaxic response to light, collect nauplii with netting or screen. Rinse with running seawater for 1–2 minutes. Dip in 400 ppm formalin for 30–60 seconds. Dip in iodophor (0.1 ppm iodine) for 1 minute. Rinse in running seawater 3–5 minutes. Transfer to disinfected larval rearing tanks.

b) *Disinfection* of tanks, equipment, pipes, air stones, etc.

For routine sanitation, hatchery and broodstock tanks (i.e. tanks for broodstock maturation, matting, spawning, larval rearing and indoor nursery) should be cleaned, disinfected and dried between use. Tanks used for the above-named purposes in crustacean (especially shrimp) hatcheries are typically precast fiberglass tanks or they are constructed of concrete or wood and either coated or painted with resin-based coatings (e.g. epoxy or fiberglass resin) or lined with plastic liners manufactured for that purpose. After harvest of the stock from the tank, all loose objects and large-sized organic debris such as algae, faeces and left-over feed should be removed. With relatively small tanks, it is advisable after harvest of the stock to fill the tank to capacity, immerse all nonporous corrosion resistant equipment (i.e.

1 Fertilised eggs are more sensitive than nauplii to formalin.

2 Nauplii are much easier to collect than are fertilised eggs in hatcheries.

airlines, air stones, stand pipes, screens, sampling containers, etc.) in the tank, and then add calcium hypochlorite to provide a minimum of 200 ppm of free chlorine. This should be allowed to set overnight. After the proper chlorinated soak-time, the tank can be drained and freshwater rinsed. Before draining the system, the treated water should be dechlorinated (see specific sections on chlorination described in this Article), unless appropriate effluent collection and treatment systems are in place. After the tank has been rinsed it should be allowed to completely dry. In the case of large tanks, an initial cleaning to remove loose debris should be followed by *disinfection* with a concentrated (~1600 ppm as chlorine) solution of calcium hypochlorite. All inside and outside surfaces should then be sprayed with this chlorine solution. The tank should then be allowed to set for several hours and then rinsed, filled and flushed. Surfaces should then be scrubbed free of all remaining material. After *disinfection* with chlorine, small or large tanks should be rinsed with clean water, then filled and flushed to ensure that no chlorine residues remain before the tank is restocked for another crop.

3. *Disinfection* of growout ponds

Following the routine harvest of a crop from a growout pond (or from a large tank or raceway used for growout of a crop), the pond (tank) bottom should be inspected. Large deposits of organic debris should be treated or removed. This is easily accomplished in lined tanks, raceways, or ponds (i.e. by flushing with a high pressure hose), but poses more of a challenge in large earth bottom ponds. Many methods of pond bottom disinfection and treatment between crops are practiced. These methods are given in detail in a number of shrimp farming manuals, and some will be listed here only with minimal details:

a) Chlorination

This *disinfectant* may be used for routine treatment of ponds between crops or when disease eradication is the goal. After draining the pond, remove (and dispose of [see section on carcass disposal under Article 5.2.4.7.c.]) as many animals from the system as is possible (this may be difficult in pond systems where the removal of large numbers of dead shrimp would not be practical). Partially refill the pond (or fill to capacity if required), discontinue the addition of new water, stop the discharge of effluent water, and remove any internal or external sources of aeration or aeration devices, which might be subject to corrosion. Then evenly distribute sufficient granulated calcium hypochlorite (such as Olin HTH™) to provide a minimum residual free chlorine concentration of 10 ppm within the entire system s water. **(NB: The person(s) applying the chlorine should wear waterproof outer ware to protect their skin, an approved chlorine mask, and goggles or a face shield for eye protection.)** Redistribute additional calcium hypochlorite as often as required to maintain the residual concentration at near or 10 ppm. Allow the system to set for a minimum of 24–48 hours (especially if applied to large systems) at this minimal chlorine concentration. The chlorine will kill all shrimp and most, if not all, of the other organisms occupying the water column or resident in the pond. After the pond has been treated with chlorine for the required minimum time and before any water is discharged, neutralise the chlorine either passively by exposure to sunlight and air for approximately an additional 48 hours (without the addition of new chlorine) or by the addition of sodium thiosulphate at a rate of five (5) molecules of sodium thiosulphate for each four (4) molecules of chlorine (or the weight of sodium thiosulphate being 2.85 times the weight of chlorine in the water, see example table below).

Pond size	Average depth	Volume	Chlorine dose	Chlorine required	HTH (65% active Cl)	Thiosulphate required
1 hectare	1 m	10,000 m^3	10 ppm	100 kg	154 kg	285 kg

Periodic testing should be done for residual chlorine; water should not be discharged until it has reached 0 ppm. Once the chlorine levels have been ascertained to be at 0 ppm, the system water can be safely dumped into the farm's effluent system. In some culture systems, in particular raceways, tanks and small lined ponds (i.e. those systems in which the majority of the shrimp were not removed prior to *disinfection*), the dead shrimp should be collected for proper disposal (see section on carcass disposal under Article 5.2.4.6.).

b) Liming

The lime, as calcium oxide or calcium hydroxide, should be applied to a very moist bottom at a rate of 5000 kg/ha or 1500 kg/ha, respectively. Great care should be taken to assure that the lime is spread evenly over the soil surface. The pond should then be allowed to set for at least a week, or at least until the soil has dried to the point of cracking to a depth of approximately 10–20 cm. Additional lime may be applied after ploughing (see below) at a rate of 50% of that normally prescribed. The pond should again be dried for at least a week, depending on the weather.

c) Drying and ploughing

Whether or not a pond is treated by chlorination or liming or left to dry untreated, ploughing is a commonly used method of treating a pond bottom to reduce its organic content, improve nutrient recycling, buffer pH, eliminate pests, and achieve *disinfection* through a combination of microbial degradation, exposure to sunlight, aeration, and desiccation. In some regions, drying and ploughing of dry pond bottoms may only be possible during the 'dry season'. When pond drying is an option, the pond bottom should be allowed to dry until the surface has cracked to a depth of approximately 10 cm. Once this level of drying has been reached, the soil should be broken up to a depth of approximately 20 cm with a plough, tiller, disk harrow, tine harrow or other similar farm implement. Ponds treated in this manner should be left for at least a week before being refilled and restocked.

4. *Disinfection* of source water

Because several of the listed *diseases* of shrimp listed in the *Code*, as well as a number of other important *diseases*, can be introduced into shrimp farms with source water when it contains vectors or carriers (i.e. wild infected crab or shrimp larvae), most farms operate with biosecurity plans that include provisions for the *disinfection* of source water. This may be accomplished by a variety of means which may include one or some combination of the following strategies:

a) Filtration of source water – source water is pumped into a supply/settling canal where it first passes through coarse bar screens to remove large aquatic animals and debris. Then the water is passed through a series of progressively finer screens, and final filtration is accomplished by passing source water through a fine mesh (200–250 µm mesh size) bag screen before being introduced into a culture pond or storage reservoir.

b) Instead of using mesh nets, some farms place filtration structures in the supply canal system. A series of compartments within these structures are filled with filter matrixes, beginning with coarse gravel for initial removal of large aquatic animals and debris, an intermediate section which contains a finer matrix of sand and gravel, and the end section which contains fine sand.

c) Chlorination and de-chlorination – Source water is pumped to a supply canal or directly into culture ponds or reservoirs (with or without filtration) and treated with sufficient chlorine to kill any potential vectors or carriers in the source water.

d) 'Zero' or reduced water exchange: Some farms use supplemental aeration and re-circulation of water in culture ponds and within the supply and discharge systems of the shrimp farm to reduce source water requirements. This reduces the volume of source water that must be disinfected before use, as well as reducing nutrient loss from farms with effluent.

Article 5.2.4.7.

Disease eradication and total facility clean-up

This action may be necessary for disease control when significant, untreatable *diseases* occur at sites where eradication is an option. The confirmed diagnosis of an listed diseases, or of an important but unlisted *emerging disease* occurring for the first time at a particular farm, at a particular site (i.e. at a quarantine facility), or within a region or country previously believed to be free of that *disease*, are events wherein it may be advisable, if not required, to eradicate the *disease* by depopulating the affected facility and performing a thorough *disinfection* of all or part of the facility.

Fallowing of the affected facility for a defined period of time may be warranted in some situations (see Chapter 1.7.1, Fallowing of Sites in this *Code*).

The following steps/actions may be used to achieve eradication of a *disease* through a total facility clean-up (TCU):

1. Depopulate all living shrimp stocks from the affected facility

   a) Discontinue stocking of the facility.

   b) Harvest and sell (if permitted) marketable stocks through normal market channels. In some circumstances cooking the product before marketing may be advisable. Cooking, either by steam or boiling water, will kill or deactivate all known disease agents of shrimp.

   c) For unmarketable stocks the following are options for disposable after harvest:

      i) Incineration: burn collected shrimp in a government approved (if required) incinerator. The limitations to this procedure are inherent to the disposal of shrimp. That is, shrimp contain large amounts of water and therefore this procedure may only be feasible for small quantities of shrimp or to larger quantities if the shrimp have been dried prior to incineration.

      ii) Burial: although this technique should be applicable to a greater number of instances, it still has its limitations. The shrimp should be placed in a pit of sufficient depth to accommodate all of the shrimp and still provide for at least 50 cm of fill covering the shrimp. The pit should be located some distance from the facility undergoing TCU and a comparable distance from any other facility culturing shrimp. Drainage from the

pit area should not be into the aquifer from which the TCU site (or any shrimp culture site) may pump its source water or into the area (estuary or beach) from which source water is drawn. Once a proper site has been selected, then the actual burial can take place. The bottom of the pit should be covered with calcium oxide (quicklime) at a rate of approximately 500 g/m^2 (5000 kg/ha) or with calcium hydroxide (slaked or hydrated lime) at a rate of approximately 150 g/m^2 (1500 kg/ha). The shrimp should be placed in the pit in layers of approximately 10 cm depth, each covered by a quantity of slaked lime or quicklime sufficient to completely cover the layer (equivalent to approximately 33–100% of the weight of the shrimp). The entire pit, including the top layer of shrimp carcasses, should then be overlaid with a minimum of 50 cm of fill dirt. In some locations, local environmental, public health and zoning officials should be consulted before the shrimp burial pit(s) is (are) dug.

2. *Disinfection* of culture tanks and ponds

See appropriate sections under Article 5.2.4.5. for methods.

3. Clean-up procedures for facility components other than culture areas

In order for a TCU to be effective, it may be necessary to disinfect the entire facility after all the shrimp have either been harvested or disposed of in some other manner. After depopulation of the facility, every possible animate and inanimate carrier of the *disease agent* must be identified and either removed from the facility or thoroughly disinfected. The movement of *disease agents* between live shrimp or dead numerous shrimp can be easily understood, while the same can not be said for their movement via inanimate components. Hence, all areas, units, subunits or components which are contaminated or potentially contaminated must go through a cleaning and disinfection process. See section 1 of Article 5.2.4.5. and Appendix 5.2.2 for a list of *disinfectants* and their methods of application.

a) Buildings

The disinfection regime used should be building-specific and dependent upon the use-pattern of that particular building.

i) Office buildings: these buildings would most often be subject only to foot traffic from people who have been in contaminated buildings or culture areas. Because of this, the greatest focus of attention should be the floors and cold storage units in the building. Floors should be thoroughly cleaned (if they are non-porous) with standard detergents and cleaning solutions, followed by a thorough drying. If the floors are carpeted, they should be vacuumed and cleaned with a detergent appropriate for carpets, or 'steam' cleaned. All other areas within these buildings, such as walls, bathrooms, desks, refrigerators, freezers, etc. should be examined for possibly contaminated materials (i.e. frozen shrimp in freezers) and any such item found and its container should be cleaned and disinfected or disposed of in a sanitary manner.

ii) Culture buildings: It must be assumed that these buildings have had direct contact with the disease agents and will therefore be handled in a different manner from that of the office buildings. The disinfection regime for these buildings will consist of two steps. First, the building should be thoroughly swept and/or vacuumed (where appropriate) to remove as much large-sized organic and inorganic debris as possible. This should be followed with the second step, treatment with chlorine. Chlorine solution (~1600 ppm) should be applied (by spraying) to all surfaces which are not

prone to the corrosive actions of chlorine. Those surfaces which should not be chlorinated, can first be sponged with a iodophor solution minimally providing 200 ppm of free iodine. These can then be covered with plastic or any other protective material. Floor surfaces can be soak-chlorinated to a depth of 5 cm with a 200 ppm chlorine solution. This should be allowed to set for a minimum of 48 hours. If many of the sprayed surfaces are somewhat susceptible to corrosion by chlorine, those surfaces can be freshwater-rinsed after the 48-hour treatment.

In buildings where *disinfection* with chlorine is not practical, fumigation with formaldehyde gas should be considered. After a general cleaning, fumigation of a sealable building can be initiated. The entire process, from the time the building is first gassed until it can be occupied again, should take a minimum of 36–60 hours. The entire building must be totally sealed off during the actual fumigation; there should be no means by which the gas can escape once it is placed in the building. If possible, the electrical service for the building should be turned off. The required environment for formaldehyde gas disinfection is a minimum temperature of 18ºC with a high relative humidity (at saturation is best, i.e. floors should be wet, etc.). Generation of formaldehyde gas is accomplished by the addition of 17.5 g potassium permanganate to each 35 ml of 100% formalin (a 37–39% aqueous solution of formaldehyde gas) for each 2.83 m^3 (100 ft^3) of space. Ideally, each room in the structure should have its own source of formaldehyde gas to assure that all areas of the building are uniformly treated. The correct amount of each compound (potassium permanganate and formalin) should be weighed out into separate containers, the formalin should be placed in a non-plastic container that is at least **10 times** the combined volume of both the formalin and the potassium permanganate. **(The person applying a formaldehyde gas fumigation should wear waterproof outer ware to protect their skin, an approved formaldehyde gas mask, and goggles or a face shield for eye protection.)** The containers with the proper amounts of the two reagents should then be placed on the floor in the centre of the room, on a large disposable protective (plastic) mat. The formalin and potassium permanganate should not be mixed at this time. Once all rooms have the correct amounts of the two compounds, the building has been completely sealed and the environment modified as necessary, the actual fumigation can begin. The mixing of the two compounds must be done very rapidly and carefully as the reaction is immediate and somewhat violent as formaldehyde gas is emitted. Starting with the room farthest from the exterior door, add the permanganate to the formalin and proceed to the next room. After all rooms have been completed, lock the exterior door and seal it from the outside with tape. The building should be allowed to set for a minimum of 12 hours. After this disinfection period the building should be flushed with clean air for 24–48 hours. There should be no detectable odour of formaldehyde when people are allowed to reoccupy the building.

An alternate method for the generation of formaldehyde gas is the sublimation of powdered paraformaldehyde. For each 2.83 m^3 (100 ft^3) of space, approximately 28 g paraformaldehyde should be used. It can be sublimated by being placed in an electric fry pan, which has been set on high. This procedure is somewhat more dangerous, because formaldehyde is flammable and a spark from such a heating device could theoretically ignite the gas. The same procedures noted above for the formalin/permanganate mixture in regards to venting, etc. should also be followed for the use of paraformaldehyde.

iii) Processing buildings: these buildings are typically constructed to permit routine

disinfection. For the most part, the procedures followed in the routine operation of such buildings are appropriate for a TCU, provided that the building, its cold rooms, and its freezers are also disinfected and thoroughly dried. If considered necessary, fumigation with formaldehyde gas may be done to insure destruction of the *disease agent(s)* of concern.

iv) Other buildings: buildings (feed storage, maintenance, tool rooms, etc.) should be treated somewhat like the office building. Care should be taken to remove all the large-sized debris, which would normally be found in relative abundance within these types of buildings. Potentially contaminated surfaces within such buildings should next be spray-chlorinated and allowed to set for 24–48 hours. This should be followed by a freshwater rinse. All equipment, which should not be exposed to the corrosive action of chlorine, should be removed before the spraying, and they should be disinfected by surface disinfection with 200 ppm of iodophor. Once the equipment has been disinfected, it can be brought back into the building. Fumigation with formaldehyde gas is another option for this type of building.

b) Culture support equipment and systems

These are operational units of the shrimp culture facility which may be housed in a building.

i) Artemia systems: All Artemia decapsulation and cyst hatching units and tanks should be treated in the same manner as other tanks. They should be cleaned of all large debris, then filled to the top with clean water and calcium hypochlorite added to achieve a final concentration of 200 ppm (free Cl_2). Chlorination should be allowed to continue for 24–48 hours. The outside of such tanks may be spray-chlorinated (1600 ppm chlorine). Treated tanks can then be dechlorinated with sodium thiosulphate, drained, freshwater rinsed, and allowed to dry for a minimum of one week. Unopened containers of Artemia cysts at the facility can be retained. These should, however, be surface disinfected with chlorine (200 ppm) or iodophor (200 ppm).

ii) Algae systems: Containers, tanks, incubators and rooms used to produce algae for feeding the larval stages of shrimp may be handled and disinfected in nearly the same way as other tanks systems. The only major difference being that special care must be taken to assure that all chlorine residues have been rinsed from the units before they are used again. In the case of the culture tubes, flasks, carboys, and flasks used to culture algae, a combination of acid (10% HCl) rinse or steam sterilisation can be used in lieu of *disinfection* with chlorine or idophor.

Disinfection of stock cultures of living algae is not possible. The use of *disinfection* is clearly out of the question; any compound which would kill the disease agent would likewise kill the algae. Hence, there are two basic methods of minimising the chance of a disease agent being present in the stock cultures.

– Dilution: all stock cultures can be cloned from the existing stocks. Each culture should be diluted either by means of serial dilutions (for broth cultures) or streaked for single colonies (agar cultures). All dilutions must be performed using strict aseptic techniques with all media being properly autoclaved. Passages from the stock cultures should not occur until the algae culture room has itself been disinfected as per the above building procedures. Once a culture has been diluted and cloned by either of these methods, to the point where there remains only

one cell of the original culture, the risk is negligible that a (shrimp) disease agent may be present.

– New Stock Cultures: If existing stock culture are discarded in a TCU, new stocks should be purchased from algae supply laboratories, or obtained from other sources where contamination with (shrimp) disease agents is unlikely, such as isolating desired species from wild populations of algae. New stock cultures should not be obtained from any facility that also cultures shrimp and may be contaminated with (shrimp) disease agents of concern.

iii) Farm equipment: Nets, seines, porous air-line tubing, etc. which are relatively inexpensive and easily obtainable should be discarded and removed from the facility during a TCU rather than being disinfected as they are not readily disinfected and chlorine is likely to damage them and shorten their useful life.

Non-expendable equipment such as large size flexible plastic tubing, pumps and pipes, transfer tanks, cages, harvest cages, harvest tables, Secchi disks, laboratory glassware, etc. should be soak-chlorinated in 200 ppm solutions for 24–48 hours. This is most easily accomplished by placing these objects in the tanks that are filled with 200 ppm solutions of chlorine. Care should be taken to have all items completely submerged (use heavy items to weigh-down more buoyant objects). A good guide is to place everything (except those that are to be thrown away) that is loose or can be unsecured from its point of attachment, into the 200 ppm chlorine solution in their respective tanks.

In the case of those similar type items which are associated with ponds, they should be placed in a special series of tanks set up near their respective ponds. These tanks should be filled with 200 ppm chlorine solutions. Following soak-chlorination, these items should be allowed to dry and be exposed to natural UV (sunlight) sterilisation. They should be turned at least once to expose all areas of the items to direct sunlight.

Tools and machinery, such as tractors, trucks, portable and stationary power tools, etc., should be thoroughly cleaned with standard cleaning solutions. All traces of mud, shrimp feed, etc. must be removed from these items. Following this, *disinfection* of surfaces likely to have been contaminated in normal use should be rinsed off with an iodophor solution (at a concentration of 200 ppm) or cleaned with steam.

Small tools and instruments such as, scales and balances, test instruments, small power tools, etc., should be gently sponged off with 200 ppm of chlorine solutions if they are inert plastic or 200 ppm of iodophor if they are otherwise. These should then be placed back in their respective buildings during the formaldehyde fumigation. High precision electronic test equipment should not be subjected to the fumigation, especially if there has been little chance that it was ever contaminated.

iv) 'New-Water' Plumbing: All new-water plumbing which is contained within buildings, especially those which have blind ends or terminate in manifolds, should be filled with a minimum 200 ppm chlorine solution. The chlorine solution should be held in the lines for 24–48 hours minimum, followed by clean water rinsing. Pipes may also be disinfected by recirculating hot water (>60ºC) through them for several hours.

v) Uniforms, boots, etc.: All items worn or used by employees should be either disposed of or thoroughly washed and disinfected. In the case of clothing, such as coveralls,

normal washing which incorporates a chlorine bleach is sufficient, especially if accompanied by sun drying. Other items, such as boots, gloves and other non-cloth items can be safely soak-chlorinated in a 200 ppm chlorine solution. This should be followed by a freshwater rinse. These items should also be contained within their respective buildings during formaldehyde fumigation.

vi) Feed items: All feed items, such as prepared feeds, fresh feeds (i.e. squid, bloodworms, frozen Artemia, bivalve mollucs, etc.) should be removed from the facility and replaced with new feeds from sources known to be free of contamination by shrimp disease-causing agents.

Article 5.2.4.8.

Re-stocking of disinfected farms

Following a TCU, restocking of the disinfected facilities or farms should be accomplished only with stocks known to be free of the *diseases* listed in the *Code* or other *emerging* or significant *diseases* of concern.

[1. Decontamination of virus in ponds and in material may be achieved by treating the surfaces with 50 parts per million (ppm) sodium or calcium hypochloride.

2. Prevention of monodon baculovirus and *Baculovirus penaei* infections in hatcheries may be achieved by prior washing of nauplii or fertilised eggs with formalin, iodophore and filtered clean seawater as described in the following figure.

		a) Nauplii*			
	Collection of nauplii using plankton net	→	Running sea water for 1– 2 minutes	→	Formalin 400 ppm for 30 seconds to 1 minute
→	Iodophore 0.1 ppm iodine for 1 minute	→	Running sea water for 3– 5 minutes	→	Hatchery ponds
		b) Fertilised eggs**			
	Collection of fertilised eggs	→	Running sea water for 1– 2 minutes	→	Formalin 100 ppm for 1 minute
→	Iodophore 0.1 ppm iodine for 1 minute	→	Running sea water for 3– 5 minutes	→	Hatchery ponds

* Nauplii are much easier to collect than are fertilised eggs in hatcheries.
** Fertilised eggs are more sensitive than nauplii to formalin.

3. Prevention of infection by infectious hypodermal and hematopoietic necrosis virus may be achieved by using specific pathogen free crustacean populations. Although this approach has proven to be useful, it is still in the experimental phase.]

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